## Photophysiology of symbiosis between haptophyte host and UCYN-A diazotroph

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The diazotrophic cyanobacterium UCYN-A shows unusual degree of metabolic streamlining<sup>1</sup> suggesting obligate symbiosis with its haptophyte host *Braarudosphaera bigelowii*<sup>2</sup>. Here we report results of the laboratory experiment studying the metabolic coupling between the host and UCYN-A. Laboratory cultures<sup>3</sup> were grown at 18°C under 12h Light/ 12h Dark cycles. Photosynthesis of the host was assayed by Chlorophyll variable fluorescence (FRR fluorometery) and by oxygen production (Clark electrode). Nitrogen fixation of UCYN-A was measured as acetylene reduction by GC.

 $N_2$  fixation in UCYN-A occurs only during the light period and is strictly light dependent and stops immediately when cells are transferred into dark. The light-dependent rate of  $N_2$  fixation in UCYN-A saturates at light intensities of 50-70 umol quanta.m<sup>-2</sup>.s<sup>-1</sup>, independently of the growth irradiance. This is significantly lower than the saturation of the photosynthesis of the host (120-250 umol quanta.m<sup>-2</sup>.s<sup>-1</sup>, depending on the growth irradiance).  $N_2$  fixation of the symbiont seems to be under circadian rhythm, cells do not fix  $N_2$  when exposed to light during the subjective night period. The light-dependent  $N_2$  fixation of the symbiont continues for several hours even if the photosynthesis of the host is fully inhibited by DCMU. This indicates that there exists a pool of reduced carbon produced by the host that fuels  $N_2$  fixation even when its photosynthesis is inhibited. On the other hand,  $N_2$  fixation can be inhibited by compounds that block thylakoid membrane electron flow in the symbiont or that collapse the transmembrane gradient. Our results confirm the active and indispensable role of Photosystem 1 in supplying reductant and/or ATP to  $N_2$  fixation in UCYN-A.

## References

- 1. Zehr, J. P., Bench, S. R., Carter, B. J., Hewson, I., Niazi, F., Shi, T., Tripp, H. J. & Affourtit, J. P. (2008) Science, 322, 1110-2.
- 2. Hagino, K., Onuma, R., Kawachi, M. & Horiguchi, T. (2013) Plos One 8.
- 3. Suzuki, S., Kawachi, M., Tsukakoshi, C., Nakamura, A., Hagino, K., Inouye, I. & Ishida, K. I. (2021) Front Plant Sci, 12, 749895